focusing uses low conductivity fluids, one can adapt the present method for use in much larger scale geometries than the micron-sized channels and capillaries described in detail herein.

Further, the previously described method can be adapted for use in modified capillary fluidic systems known to one of ordinary skill in the art. FIG. **6** depicts a capillary fluidic system having a capillary tube **70** spanning between two fluid reservoirs **77**. Two temperature blocks, denoted as heated block **72** and cooling block **73** are located along the length of the capillary tube **70** to provide a desired temperature gradient in the capillary tube **70**. Alternatively, temperature blocks being both cooling, both heated, both at ambient temperature, or any combination, thereof, may be substituted to provide the desired temperature gradient.

The fluid reservoirs **77** contain a fluid with temperature dependent ionic strength. Electrodes **74**, **75** are connected at one end to a power supply and on the other end, are in contact with the fluid in the reservoirs **77**. The power supply applies a driving voltage through the capillary tube **70**. A source of bulk flow is driven either by electro-osmosis with the applied driving voltage, by a pressure gradient applied, e.g. by a pump, or a combination of the two. Detector **80** is used to detect materials present in the fluid.

The temperature gradient focusing technique of the present invention can be used for mixing reactions and for monitoring and/or detecting the interactions between different materials such as interacting molecular species. In one form of mixing reactions/interactions, a material is first focused using temperature gradient focusing as described above with regard to the prior embodiments. Subsequently, a second material is introduced into the fluid conduit and a product of the interaction of the two materials can be focused on the same temperature gradient and detected. The materials and/or their interaction products can be detected in accordance with conventional detection methods known to one of ordinary skill in the art as well as in accordance with previously described detection methods.

Referring now to FIGS. **7**(*a*) and **7**(*b*), mixing reactions in accordance with the present invention can be conducted using a fluid conduit such as microchannel **710** which comprises a hot zone and a cold zone produced by heated block **718** and cooled block **720**, respectively. This creates a temperature gradient in the fluid **716** which contains a first material to be separated or focused. An electric field is established in the fluid **716** by applying a voltage potential to a high voltage electrode **712** so that the voltage between terminal **712** and a grounding electrode **714** causes the first material to move electrophoretically. The voltage is most commonly applied by an electrical power supply **726**.

As with the prior embodiments, a temperature gradient is established along the length of the microchannel **710** which has a significant component substantially aligned with the electric field to thereby generate a gradient of the electrophoretic velocity of the first material. The gradient of electrophoretic velocity of the first material can be established by the fluid **716** having an ionic strength or pH which is temperature dependent as described above.

A bulk flow is produced in the fluid **716** due to electroosmosis resulting from the applied voltage and adjusted via pressure controller **728** to have a significant component substantially aligned in the direction opposite of the direction of the electrophoretic migration of the first material so that the total velocity of the first material is equal to zero at a position along the microchannel and a focused band of material F_1 will form at that position. The bulk flow is established in a similar manner to that described with regard to the prior embodiments.

Next, a second material is introduced into the fluid so as to move through the now focused first material band F_1, and to interact with the first material. The second material can be introduced to either end of the microchannel, depending on whether it has an electrophoretic mobility that is less than or greater than that of the first material. If the electrophoretic mobility of the second material is less than that of the first material, or if the second material is of the opposite charge than the first material, then the second material should be introduced into the end of the channel where the bulk fluid flow predominantly determines the direction of motion of the first material. More specifically, if a fluid with a temperature-dependent ionic strength is used, the second material should be introduced into the high ionic strength end. If, on the other hand, the second material has an electrophoretic mobility that is greater than that of the first material, assuming that they have the same sign of charge, it should be introduced into the end of the channel where the electrophoretic motion dominates or the low ionic strength end.

After the second material is introduced, if hybridization, binding, or other interaction or chemical reaction of the two materials occurs, the product of that interaction, i.e., a third material, will be focused at a second position and a second band F_2 will be observed as depicted in FIG. **7**(*b*).

Preferably, the position of the second band F_2 will be different than the position of the first band F_1, due to a difference in electrophoretic mobility.

In a non-limiting example of this method, this method can be used to observe the interaction of single strand DNA with another nucleic acid. In this example, the first material is negatively charged single stranded (ss) DNA which is labeled for observation within the microchannel **710**, and the second material is a peptide nucleic acid (PNA) which is introduced into the microchannel **710**. First, the ssDNA is focused as a single band F**1**. Then, the neutral PNA is introduced into the microchannel **710**. The PNA is carried by the bulk flow of the fluid through the stationary, focused band of single strand DNA at location F**1**.

The ssDNA and PNA are allowed to interact with each other. If the PNA and ssDNA hybridize, the hybrid duplex will focus at a different spatial location within microchannel **710** because of its different electrophoretic mobility and as a result a second band, F_2, is observed. If the PNA is not complimentary to the focused ssDNA, the two will not hybridize with one another and the neutral PNA will remain unfocused. In such an instance, a second band will not be observed and the result will be the same as shown in FIG. **7**(*a*).

The following non-limiting example is included to provide further understanding of the present mixing reaction method using temperature gradient focusing. The ssDNA was fluorescently labeled on the 5' end while the PNA probes contained a fluorescence label on the N-Terminus. The labels were fluorescein for DNA (green emission) and TAMRA for PNA (orange emission). Using a VHS tape and frame grabbing software, the hybridization of a perfect complement PNA probe to ssDNA was recorded. The experiment was performed in a 30 μm I.D. fused silica capillary, 10 cm long. The capillary was mechanically and thermally anchored to two copper blocks at different temperatures to create a temperature gradient along a 2 mm section approximately midway along the length of the capillary. A temperature gradient from 10° C. to 80° C. was applied. The fluid used was 0.1 mol/L Tris(hydroxymethyl)aminomethane, 0.1 mol/L phenol